

US 10/058,568

Amended claims - clean copy



What is claimed is:

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1. An isolated nucleotide sequence comprising the BS92-7 gene.
 2. An isolated nucleotide sequence that mediates male fertility in plants comprising a nucleotide sequence encoding any of the amino acid sequences of SEQ ID Nos. 2, or 8 and those sequences which hybridize to the nucleotide sequences encoding any of the amino acid sequences of SEQ ID Nos. 2, or 8 under highly stringent conditions.
 3. An isolated DNA molecule that mediates fertility in plants comprising a nucleotide sequence of any of SEQ ID Nos. 1, 3, or 7 and those sequences which hybridize to the nucleotide sequences of SEQ ID Nos. 1, 3, or 7 under highly stringent conditions.
 4. A plant cell transformed by the nucleotide sequence of Claim 1.
 5. A plant cell transformed by the nucleotide sequence of Claim 2.
 6. A plant cell transformed by the nucleotide sequence of Claim 3.
 7. A plant transformed by a nucleotide sequence of Claim 1.
 8. A plant transformed by a nucleotide sequence of Claim 2.
 9. A plant transformed by a nucleotide sequence of Claim 3.
 10. The plant cell of Claim 4 wherein the cell is a maize cell.
 11. The plant of Claim 7 wherein the plant is maize.
 12. A method of impacting fertility of a plant comprising impacting the BS92-7 gene.
 13. A method of impacting fertility of a plant comprising impacting a nucleotide sequence in the plant encoding the amino acid sequence of any of SEQ ID Nos 2, or 8, the nucleotide sequences of any of SEQ. ID Nos. 1, 3, or 7 and those sequences which hybridize to any of said sequences under highly stringent conditions.
 14. The method of Claim 12 wherein the sequence expression is repressed.
 15. The method of Claim 12 wherein expression of the nucleotide sequence is repressed by mutation of the nucleotide sequence.
 16. The method of Claim 12 further comprising delivering into the plant a second nucleotide sequence which represses expression of the nucleotide sequence.
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17. The method of Claim 16 further comprising delivering into the plant a second nucleotide sequence molecule oriented in the antisense direction relative to the DNA molecule thereby repressing expression of the DNA molecule.
18. The method of Claim 12 wherein a native BS92-7 gene in a plant is silenced and a second BS92-7 gene linked to an inducible promoter is introduced into the plant such that the plant is constitutively male sterile and fertility is induced by inducing the promoter.
19. A method of producing hybrid seed, comprising: (a) planting in cross-pollinating juxtaposition, a first seed from a selected male fertile parent line and a second seed selected from a female parent line having male sterility produced according to the method of Claim 12; (b) growing the seed to mature plants under conditions which do not induce expression of the second DNA molecule; (c) cross-pollinating the male sterile female plant with pollen from the male fertile plant; and (d) harvesting seed from the male sterile female plant.
20. The method of claim 19 further comprising cross-fertilizing the male sterile plant with a second plant, the second plant comprising a second exogenous gene, the product of the second gene preventing disruption of the male tissue by the first exogenous gene, producing a male fertile hybrid plant.
21. The method of claim 19 wherein the gene impacting male fertility is dominant and further comprising growing the hybrid seed to produce a third male sterile parent plant; producing a fourth parent plant comprising one or more genes controlling a desired gene trait and cross-fertilizing the third and fourth parent plants to produce second hybrid seed.
22. A male fertility mediated plant produced according to the method of Claim 12.
23. A method of providing heritable externally controllable male sterility in a plant comprising linking BS92-7 in an expression sequence with an inducible promoter responsive to external control; delivering the expression sequence into the genome of the plant; and inactivating a second DNA molecule which codes for the product of BS92-7 from the native genome of the plant.
24. The method of claim 23 wherein the nucleotide sequences comprise the nucleotide sequences encoding the amino acids of SEQ ID Nos. 2, or 8 or having the nucleotide

sequence of any of SEQ. ID Nos. 1, 3, or 7 and those nucleotide sequences which hybridize to any of said sequences.

25. A method of reproducing a plant having heritable, externally controllable male sterility produced according to the method of Claim 23 further comprising planting seed of the plant to provide growing male sterile plants; inducing conversion of the growing plants to male fertile form under conditions which induce the promoter to express the first DNA molecule; and open-pollinating the growing plants in isolation to produce seed; and harvesting the seed.
26. A controllably male sterile plant produced according to the method of Claim 23.
27. An expression vector comprising the DNA sequence of Claim 1.
28. The expression vector of claim 27 further comprising an exogenous gene, wherein the exogenous gene is operably linked to the promoter.
29. The expression vector of claim 27 wherein the promoter is selected from any one of CaMV35S, SGB6, BS92-7, MS45 or 5126.
30. The expression vector of claim 27 wherein the product of the exogenous gene disrupts the function of male tissue.
31. Plant cells comprising the vector of claim 27.
32. A method of mediating male fertility in a plant comprising introducing into a plant the expression vector of claim 27 wherein the exogenous gene impacts male fertility of the plant and the promoter controls expression of the exogenous gene.
33. The method of claim 32 wherein a regulatory element in conjunction with the promoter is inducible.
34. A nucleotide sequence as represented in ATCC deposit no. 98932.
35. An isolated nucleic acid sequence encoding the promoter region of the gene BS92-7.
36. An isolated nucleic acid sequence comprising SEQ ID NO. 5 or those nucleotide sequences which hybridize to SEQ ID NO. 5 under conditions of high stringency.
37. A male tissue-preferred regulatory region comprising nucleotide sequences essential for initiating transcription of the BS92-7 gene.
38. A male tissue-preferred regulatory region comprising Seq. ID. NO. 6.
39. An isolated nucleic acid that is a male tissue specific regulatory element comprising the nucleotide sequence of SEQ ID No. 6 or those nucleotide sequences which

- hybridize to SEQ ID. NO. 6 under conditions of high stringency wherein the regulatory element is essential for initiating transcription of Seq ID Nos. 1, 3 or 7.
40. The regulatory region of claim 37 comprising nucleotide sequences of about 130 contiguous base pairs upstream of the TATA box of the BS92-7 gene.
 41. A regulatory region comprising the sequences of -141 to -83 upstream of the TATA box of SEQ ID NO. 7 or those nucleotide sequences which hybridize to said sequences under conditions of high stringency
 42. The regulatory region of claim 41 comprising sequences -112 to -93 upstream of the TATA box of SEQ ID. No. 7 or those nucleotide sequences which hybridize to said sequences under conditions of high stringency.
 43. The regulatory region of claim 41 comprising sequences -161 to -152 upstream of the TATA box of SEQ. Id. No. 7 or those nucleotide sequences which hybridize to said sequences under conditions of high stringency
 44. The regulatory region of claim 41 comprising sequences -141 to -132 upstream of the TATA box of SEQ. Id. No. 7 or those nucleotide sequences which hybridize to said sequences under conditions of high stringency
 45. The regulatory region of claim 41 comprising sequences -92 to -83 upstream of the TATA box of SEQ. Id. No. 7 or those nucleotide sequences which hybridize to said sequences under conditions of high stringency
 46. An expression vector comprising a promoter that is operably linked with the male tissue specific regulatory element of claim 37.
 47. The expression vector of claim 46 further comprising an exogenous gene, wherein the exogenous gene is operably linked to the promoter.
 48. The expression vector of claim 46 wherein the promoter is selected from any one of CaMV35S, SGB6, BS92-7 MS45 or 5126.
 49. The expression vector of claim 46 wherein the product of the exogenous gene disrupts the function of male tissue.
 50. Plant cells comprising the vector of claim 46.
 51. A method of mediating male fertility in a plant comprising introducing into a plant the expression vector of claim 46 wherein the exogenous gene impacts male fertility

of the plant and the regulatory element in conjunction with the promoter control expression of the exogenous gene.

52. The method of claim 51 wherein the exogenous gene disrupts function of male tissue of the plant causing the plant to be male sterile.
53. The method of claim 52 wherein a (?) regulatory element in conjunction with the promoter is inducible.
54. The method of claim 51 wherein the plant is constitutively sterile when the promoter and regulatory element are not induced and is fertile when the promoter and regulatory element are induced.
55. The method of claim 51 further comprising cross-fertilizing the male sterile plant with a second plant, the second plant comprising a second exogenous gene, the product of the second gene preventing disruption of the male tissue by the first heterologous gene, producing a male fertile hybrid plant.
56. A method of producing hybrid seeds comprising: (a) producing a first parent plant comprising nucleotide sequences essential for initiating transcription of the BS92-7 gene operably linked with an exogenous gene impacting male fertility of the plant such that the plant is male sterile; (b) producing a second parent plant which is male fertile; (c) cross-fertilizing the first parent plant and the second parent plant to produce hybrid seeds.
57. The method of claim 56 wherein the gene impacting male fertility is dominant and further comprising growing the hybrid seed to produce a third male sterile parent plant; producing a fourth parent plant comprising one or more genes controlling a desired gene trait and cross-fertilizing the third and fourth parent plants to produce second hybrid seed.